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Molecular epidemiology of *Staphylococcus aureus* nasal colonization among patients and their parents /guardian in an Iranian referral hospital

Babak Pourakbari [a], Mahmoud Khodabandeh [b], Shima Mahmoudi [a], Farah Sabouni [b], Alireza Aziz-Ahari [c], Abbas Bahador [d], Sepideh Keshavarz Valian [e], Reihaneh Hosseinpour Sadeghi [a], Setareh Mamishi [a, b, *]

[a] Pediatric Infectious Disease Research Center, Tehran University of Medical Sciences, Tehran, Iran
[b] Department of Infectious Diseases, Pediatrics Center of Excellence, Children's Medical Center, Tehran University of Medical Sciences, Tehran, Iran
[c] Radiology Department, Rasool-e-Akram Hospital, Iran University of Medical Sciences, Tehran, Iran
[d] Department of Microbiology, Tehran University of Medical Sciences, Tehran, Iran
[e] School of Medicine, Tehran University of Medical Sciences, Tehran, Iran

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ABSTRACT

Introduction: Carriage of *Staphylococcus aureus* in the nose appears to play a key role in the epidemiology and pathogenesis of infection. It is important to investigate the genetic relatedness of *S. aureus* and MRSA clones in different geographic regions. The aim of this study was to assess the nasal carriage rate of *S. aureus*, including MRSA strains in both hospitalized children and general adult population (parents/guardian). In addition, antibiotic susceptibility pattern and molecular diversity of *S. aureus* in both population was evaluated in an Iranian referral pediatrics Hospital.

Material and methods: All samples were obtained through nasal screening of patients and general adult population at admission and discharge day. The prevalence, resistance, and molecular diversity of all *S. aureus* isolates were examined.

Results: In the current study, nasal carriage of *S. aureus* and *Staphylococcus* non *aureus* was identified in 384 (26%) and 1004 (68%) of the study population. The prevalence of MRSA nasal carriage in children and adults was 6.6% (29 out of 438) and 2.8% (29 out of 1046), respectively.

Among *S. aureus* strains isolated obtained from patients and general adult population at admission day, high sensitivity to most of the antibiotics such as vancomycin (100%), rifampin (95%), linezolid (94%), quinupristin/dalfopristin (94%), minocycline (94%), chloramphenicol (89%), gentamycin (87%), amikacin (87%), clindamycin (86%) and moxifloxacin (83%) was seen. The most resistance antibiotics were penicillin (96–98%) and methicillin (44–47%). The susceptibility patterns of nasal *S. aureus* strains isolated at discharge day was not statistically different from *S. aureus* isolates obtained at admission day. Admission *S. aureus* isolated strains of 77 patients (64%) were similar to the isolated *S. aureus* strains of discharge, while *S. aureus* isolated strains of 43 patients (36%) was not similar to the strain of discharge (had similarity of less than 70%).

Conclusion: High prevalence of nasal carriage of *S. aureus* and MRSA indicates the urgent need to improve strategies for management of *S. aureus* infections.

Our findings are useful for understanding of *S. aureus* nasal colonization dynamics within the patients and general population. Surveillance for *S. aureus* in community settings can provide data on circulating strains and might help developing control measures for reducing of infection spread in hospitals.

1. Introduction

Among hospital-acquired infection in developing countries, *Staphylococcus aureus* is the second most recognized causes after

* Corresponding author. Department of Pediatric Infectious Disease Children Medical Center Hospital School of Medicine Tehran University of Medical Sciences No.62, Gharib St., Keshavarz Blvd., Tehran, Iran.
E-mail address: smamishi@sina.tums.ac.ir (S. Mamishi).

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the *Enterobacteriaceae* [1]. *S. aureus* is one of the most common public health problem that increases the overall burden of infectious disease in each setting [2]. Carriage of *S. aureus* in the nose appears to play a key role in the epidemiology and pathogenesis of infection [3]. Nosocomial infections caused by methicillin-resistant *S. aureus* (MRSA) pose a serious problem in many countries; however, the exact burden of disease caused by MRSA remains largely unknown [4].

One of the cardinal features of the rapid emergence of MRSA in many parts of the world is the dissemination of specific clones which has contributed to the accelerated increases in the incidence of MRSA. Therefore, it is important to investigate the genotypic characteristics and evolutionary pathway of MRSA clones as well as the genetic relatedness of the strains isolated in different geographic regions [5].

Early identification of patients colonized with MRSA and subsequent prevention of its spread through infection control measures are believed to be essential [4]. It has been reported that patients colonized with MRSA on admission were 15 times more likely to develop MRSA Infection [6,7].

Identification of the epidemiology and infection patterns in local hospital settings remains critical as it directly affects clinical management plans and decision making about implementation of a national universal screening program for MRSA. The aim of this study was to evaluate the prevalence, resistance, and molecular diversity of nasal *S. aureus* colonization/infection patients and general adult population (parents/guardian) in an Iranian referral pediatrics Hospital.

2. Material and methods

2.1. Sample collection and culture

This study was performed on patients and their parents/guardian (general adult population) in an Iranian referral pediatric Hospital. In our study parents/guardians represented as general adults population; therefore, we asked them to participate in our study if they had informed consent. In some cases, only the parents/guardians were participated and their child was not taking part. All patients and their parents/guardian were required to give written informed consent to participate.

Sampling on admission and discharge consisted of swabbing of anterior nares. Admission swabbing took place within the first hours after admission. Patients with missing swabs or swabs taken outside the time criteria were excluded from analysis.

Discharge was defined as the patient leaving the hospital to another hospital or care institution or to home. Discharge screening took place up to 24 h before discharge.

The demographic information of all patients was recorded in questioner's forms.

Nasal swabs were obtained by trained examiners from both anterior nares using premoistened swab. Swabs were inserted into the nasal vestibule, and the swab was rotated four times. Trypticase soy broth (Merck, Germany) was used as the transport medium. Nose swabs were transported at 4 °C in a transportable compressor cooler and processed immediately after sampling. Samples were sent to the laboratory and were inoculated on mannitol salt agar plates and incubated at 35 °C for 48 h. For microbiological analysis, one suspicious colony was chosen from the selective agar. *S. aureus* was identified by colony morphology on mannitol salt agar plates, Gram stain, catalase test and coagulase test.

2.2. Antimicrobial susceptibility testing

A standardized Kirby-Bauer disc-diffusion method was performed on Mueller-Hinton agar according to the Clinical and Laboratory Standards Institute (CLSI) criteria [8].

Isolates were tested for susceptibility to the following antibiotics: minocycline, trimethoprim-sulfamethoxazole, quinupristin/dalfopristin, vancomycin, clindamycin, chloramphenicol, tetracycline, amikacin, ciprofloxacin, gentamycin, moxifloxacin, linezolid, rifampin, penicillin, cefoxitin, chloxacillin, azithromycin and cefazolin. The diameters of incubation halos were interpreted after 24 h of incubation at 37 °C.

2.3. Molecular testing

DNA was extracted from *S. aureus* isolates using lysostaphin digestion, as described previously [9]. Confirmation of MRSA was achieved by polymerase chain reaction (PCR) targeting the *mecA* gene [10].

2.4. ERIC-PCR reaction

S. aureus isolates were genotyped by enterobacterial repetitive intergenic consensus PCR (ERIC-PCR), as described previously [10]. The primers used were ERIC-1R (5′- ATGTAAGCTCCTGGGGATTCAC-3′) and ERIC-2 (5′- AAGTAAGTGACTGGGGTGAGCG-3′).

2.5. Data collection and analysis

All data were analyzed by SPSS version 13.5 (SPSS Inc., Chicago, IL). Continuous data were presented as mean ± standard deviation and/or median (25% quantile, 75% quantile). Categorical data were presented as percentages. Student's t-test was used for statistical analysis to compare the means between the two groups. A P value of $\leq$0.05 was considered statistically significant. Comparison of ERIC-PCR banding patterns was performed using Gelcompar II, version 6.5 (Applied Maths, Sint-Matens- Latem, Belgium). Isolates producing fingerprints showing more than 80% relatedness (Dice coefficient/unweighted pair-group method with arithmetic mean [UPGMA]) were allocated to the one ERIC-PCR type.

3. Results

3.1. Sample sources and patient and their parents/guardians demographics

A total of 1484 swabs were taken, 438 samples from children and 1046 samples from adults. In the current study, overall carriage rate nasal carriage of *S. aureus* and *Staphylococcus* non *aureus* from both pediatric children and adults was 384 (26%) and 1004 (68%), respectively. Of these, 146 children (33%) and 238 adults (23%) were found to carry *S. aureus* (Fig. 1). The mean age of the children and adults was 3.8 ± 3.6 and 29.7 ± 6.2 years, respectively.

Among 146 patients, nearly half of the patients were less than 1 year old (n = 74, 51%), while 14 cases were between 1 and 2 years (10%), 14 cases between 2 and 3 years and 44 patients were more than 3 years old (30%). The mean interval day of admission to discharge patients was 6.3 ± 4.9.

The mean interval time of hospital stay among the patients that had different admit/discharge strains was 6.8 ± 6.1 day that was not significantly different from patients with similar admit/discharge strains (5.4 ± 1.5 day) (p value > 0.05).

The hospital location was not significantly different between patients with different or similar admit/discharge strains (64% and 60% of with patients with different admit/discharge strains had been hospitalized in infectious ward and the rest of them were in gastroenterology ward, while 60% of patients with similar admit/discharge strains were hospitalized in infectious ward).